

Reprints From *Agricultural
Research* magazine

Northern Plains Area 2002 Research Highlights

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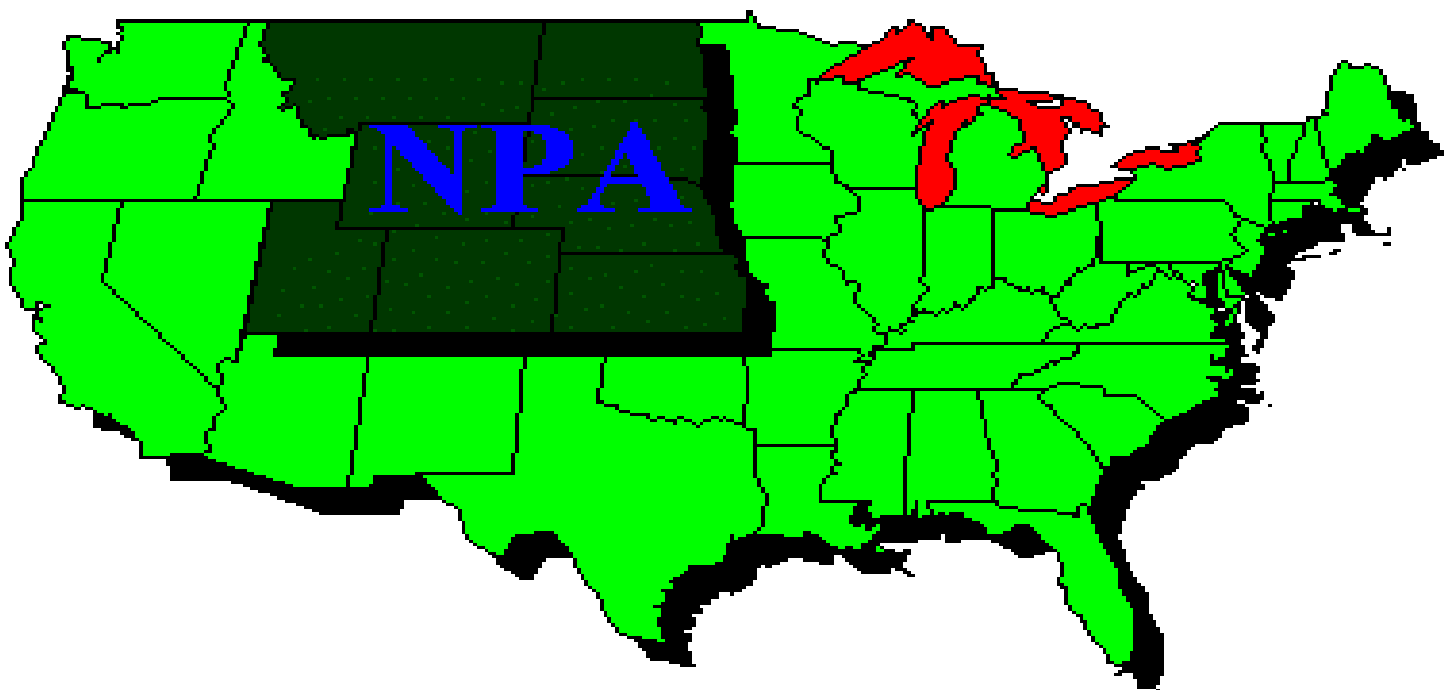
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Integrating scientific knowledge into systems that help Americans make the most of our resources and enable the transfer of technology from laboratory to farm.



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So the Rain Stays in the Plain

Crop Sequence CD Helps Farmers Fight Drought

It snowed a little on March 5 in Minot, North Dakota, close to the Canadian border. Farmers there were glad to see the snow because they know that when it melts it will provide precious water to the Northern Plains area, which is drier than usual this year.

While the situation doesn't seem as dire as it does for the eastern United States, it's still one that farmers kept in mind this past winter as they planned their spring planting.

"When we talk rotation in the Northern Great Plains," says Larry Kleingartner, executive director of the National Sunflower Association in Bismarck, North Dakota, "we're not talking corn one year and soybeans the next. We're talking about rotations in which soybean is but one among a dozen options." And some of those combinations will do better than others with less water. The trick is knowing which ones.

That's why Kleingartner and farmers in his association and elsewhere are clamoring for the Crop Sequence Calculator CD recently released by the Agricultural Research Service. It calculates crop performance with 100 combinations of 10 crops: barley, canola, crambe, dry bean, dry pea, flax, safflower, soybean, sunflower, and wheat.

The calculator uses data from several years' worth of research by ARS and others to provide producers with scientific research information on the sequencing of crops. To obtain crop-risk information for the calculator, a team of scientists at the ARS Northern Great Plains Research Laboratory in Mandan, North Dakota, grew all these combinations in 1999 and 2000 as part of a crop sequence project. The team—which includes a plant pathologist, three soil scientists, and three rangeland scientists—recorded data on crop production, plant diseases, weeds, crop water use, and amount of soil protected by crop residue.

The CD is a calculator of potential returns, a reference library, and a set of slide shows, some showing research results, all in one.

It even contains photo guides to the weeds, insects, and plant diseases likely to pose problems. And some of the slide shows

explain the basics of soil properties associated with soil quality, crop root growth and soil-water use, and pest management.

In addition to the Mandan team, Dave Archer, an ARS economist in Morris, Minnesota, Randy Anderson, an ARS weed scientist in Brookings, South Dakota, and Janet Knodel, a crop protection specialist at North Dakota State University in Minot, provided information on their areas of expertise.

Using the calculator with drought in mind, farmers would learn that planting a crop after beans or peas would be the best bet, since these use the least amount of soil water. Peas offer the best chance of leaving

the most soil water for the next crop.

And a farmer would find that planting peas before sunflowers promises the highest sunflower yield—1,490 pounds an acre. By plugging in a typical price of 9 cents a pound, the calculator would show gross earnings of \$134 an acre. By clicking on the "Production Economics" button, farmers could see an estimated average net return of \$42.41 per acre for that rotation, from the 1999 and 2000 experiments.

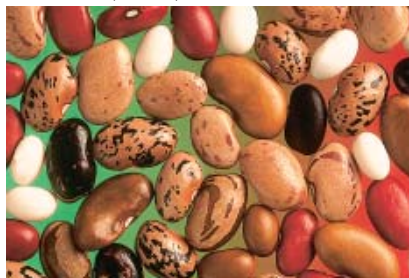
Safflower and sunflowers are the deepest rooting crops but are also among the lowest residue producers. Soil erosion could be a serious problem if low-residue crops are grown 2 years in a row. During drought years, the amount of residue is even less, which exposes the soil to greater evaporation and erosion.

The CD also lists useful web sites to go to for more information. The Crop Sequence Calculator can be ordered for free online from the Northern Great Plains Research Laboratory web site at <http://www.mandan.ars.usda.gov>.—By **Don Comis**, ARS.

This research is part of Soil Resource Management, an ARS National Program (#202) described on the World Wide Web at <http://www.nps.ars.usda.gov>.

For more information about the CD, contact Joe Krupinsky at the USDA-ARS Northern Great Plains Research Laboratory, P.O. Box 459, Mandan, ND 58554; phone (701) 667-3011, fax (701) 667-3054, e-mail krupinsj@mandan.ars.usda.gov. ♦

KEITH WELLER (K8397-15)



Beans.

BRUCE FRITZ (K5752-11)



Sunflowers.

MICHAEL THOMPSON (K7394-6)



Winter wheat.

Shortcuts to Disease-Resistant Wheats

Everyone likes to take shortcuts in time-consuming tasks. And wheat breeders are no exception. Someday, wheat breeders may be able to use new molecular tools being developed by ARS in collaboration with Kansas State University and the Kansas Wheat Commission.

These tools show promise for reducing the time it takes breeders to move important quality and resistance traits into breeding populations of wheat using conventional breeding techniques. Currently, it can take as long as 10 or more years to develop new wheat varieties.

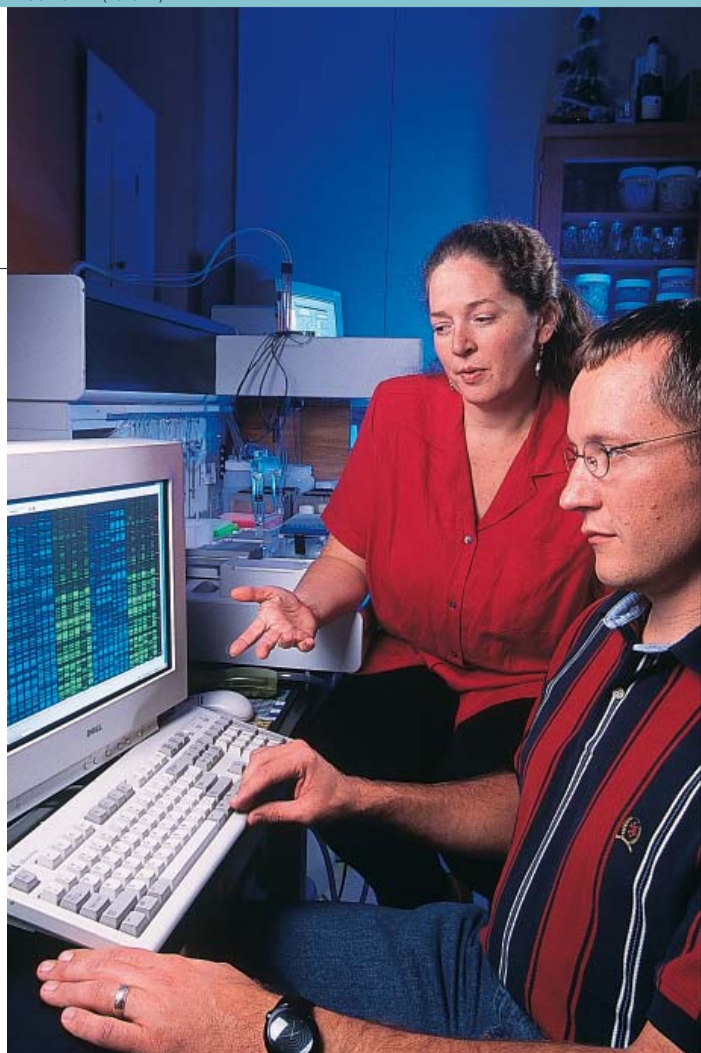
“Using molecular (or DNA) markers may shorten the task of improving insect and disease resistance while maintaining good yield and quality characteristics,” says plant geneticist Gina L. Brown-Guedira in ARS’ Plant Science and Entomology Research Unit in Manhattan, Kansas.

Molecular markers are small pieces of genetic material—DNA—that can be seen on a gel and are known to be reliably linked in this case to resistance genes. They offer breeders a fast and safe way to identify wheat resistant to pathogens.

Brown-Guedira and ARS molecular biologist John P. Fellers are focusing on finding markers that will ultimately be used to incorporate longer-lasting resistance to major wheat diseases, such as leaf rust, Karnal bunt fungus, and fusarium head scab. (See “Tagging New Leaf Rust Resistance Genes in Wheat,” *Agricultural Research*, May 2001, p. 19.)

One major accomplishment by scientists in this laboratory is identification of a molecular marker for a gene that holds the key to nearly 25 percent of the resistance to Karnal bunt fungus. This fungus is currently quarantined by 72 countries, making it a threat to our export markets. Besides yield losses, Karnal bunt disease lowers the quality of flour used for food.

Scientific studies on Karnal bunt are limited to geographic areas where the fungus is present. Working with the fungus in noninfected areas is restricted to guard against potential spread.



Geneticist Gina Brown-Guedira and molecular biologist John P. Fellers review DNA marker data from disease-resistant wheat breeding populations.

“But markers can be used at any stage of plant growth without having to infect plants with disease,” says Fellers.

So far, researchers in the United States and abroad have identified markers for disease-resistance genes, insect-resistance genes, and quality and environmental stress genes in wheat that can be applied to wheat breeding programs.—By **Linda McGraw**, formerly with ARS.

This research is part of Plant, Microbial, and Insect Genetic Resources, Genomics, and Genetic Improvement (#301) and Plant Diseases (#303), two ARS National Programs described on the World Wide Web at <http://www.nps.ars.usda.gov>.

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Fighting

Insect Pests of Stored Foods

PEGGY GREB (K9724-1)



Entomologist Mike Mullen checks pheromone traps for captured Indianmeal moths as part of a program to develop an effective monitoring system for stored-product insect pests.

PEGGY GREB (K9725-1)



Indianmeal moths trapped on a pheromone trap.

Anyone who has ever been to a picnic knows insects are drawn to food. That's why developing new methods to keep insects out of food in packages, warehouses, and processing plants is critical for food manufacturers.

New and innovative methods are needed because the industry is challenged to reduce pesticide use while ensuring that food products are insect-free. To meet these challenges, a team of ARS scientists at the Grain Marketing and Production Research Center in Manhattan, Kansas, is working closely with industry.

Keep Out, Bug!

Keeping food in containers is one of the oldest ways to protect food from insects. Ancient historical documents describe the use of crude containers, such as gourds, leaves, shells, animal skins, and even human skulls. In the 1800s, people turned to paperboard boxes, paper bags, and tin cans to preserve perishables. In the 1900s, the most popular materials for preserving food were aluminum foil, cellophane bags, and plastic.

Today, restrictions on pesticide use and having fewer sanitation personnel at various points along the distribution chain have made insect-resistant packaging even more important to consumers and to food or feed manufacturers.

Entomologist Michael A. Mullen, in cooperation with several food manufacturers, has conducted packaging studies on a variety of products, including cereals, raisins, baby foods, and dry pet foods. Mullen classifies insects as either invaders, which enter through existing openings, or penetrators, which can chew through packaging materials. (See "Pest-Proofing Food Packaging," *Agricultural Research*, March 1998, pp. 10–11.)

"Simply using a different glue pattern in the seals and closures of bags can help safeguard the product from insects. A glue pattern that forms a complete seal with no channels for insects to crawl through can help prevent insect entry into a package," says Mullen. Another method is to use tightly fitting overwraps to increase resistance to invasion.

But packaging is just one defense. "Food processors should follow good sanitation practices along with insecticide treatments," says ARS entomologist Franklin H. Arthur. In flour mills and food processing plants, insects that survive an insecticide treatment could live on food or crumbs left by poor sanitation. These surviving insects may become resistant to insecticides, making it harder to eliminate the infestation and prevent economic damage.

As an alternative to insecticides, Arthur is testing insect growth regulators (IGRs), chemicals that prevent insect larvae from becoming reproductive adults. To replicate food-storage conditions, Arthur creates "exposure arenas" by pouring concrete into petri dishes. These test arenas are used to study insect survival after exposure to IGRs and various insecticides. The chemicals are sprayed directly onto the concrete, and insects are exposed to the treated surfaces.

IGRs aren't toxic to humans, and they can suppress populations of important stored-product insect pests, such as the red flour beetle and the confused flour beetle. Arthur recently

evaluated a volatile formulation of the IGR hydroprene, known commercially as Pointsource, to control these two beetles. In laboratory tests, larvae of both beetle species exposed to Pointsource often failed to molt to the adult stage. Adult insects that did emerge were usually deformed and died quickly. Use of this product could be most effective in small, confined spaces in retail stores and homes.

Trapping the Enemy

Food products can become infested by insects during storage at any point from the manufacturer to the kitchen cupboard. Traps baited with nontoxic chemical lures called pheromones can reduce the need for insecticides by monitoring, detecting, and pinpointing insect infestations.

Mullen has developed an insect monitoring system using specially designed traps and pheromones. By establishing a grid of traps designed for crawling and flying insects and plotting the number of insects collected in each trap, he can map insect populations for facility managers. This allows precise identification of infested materials and helps target—and thus limit—use of chemical control methods.

Mullen and Alan K. Dowdy, formerly an ARS entomologist at the Grain Marketing and Production Research Center, worked cooperatively with Trécé, Inc., of Salinas, California, to develop a trap that can be hidden under shelves in retail stores, warehouses, food processing facilities, and home pantries. Commercially sold as Discreet Trap, it is expected to increase use of monitoring devices in retail areas and reduce the need for pesticides by pinpointing infestations.

Another trap, marketed by Trécé, Inc., as Dome Trap, was originally developed by Mullen. He and Oklahoma State University scientists later modified it to include a dust cover. Since dust can clog traps, making it possible for insects to escape, keeping dust out makes the traps more effective.

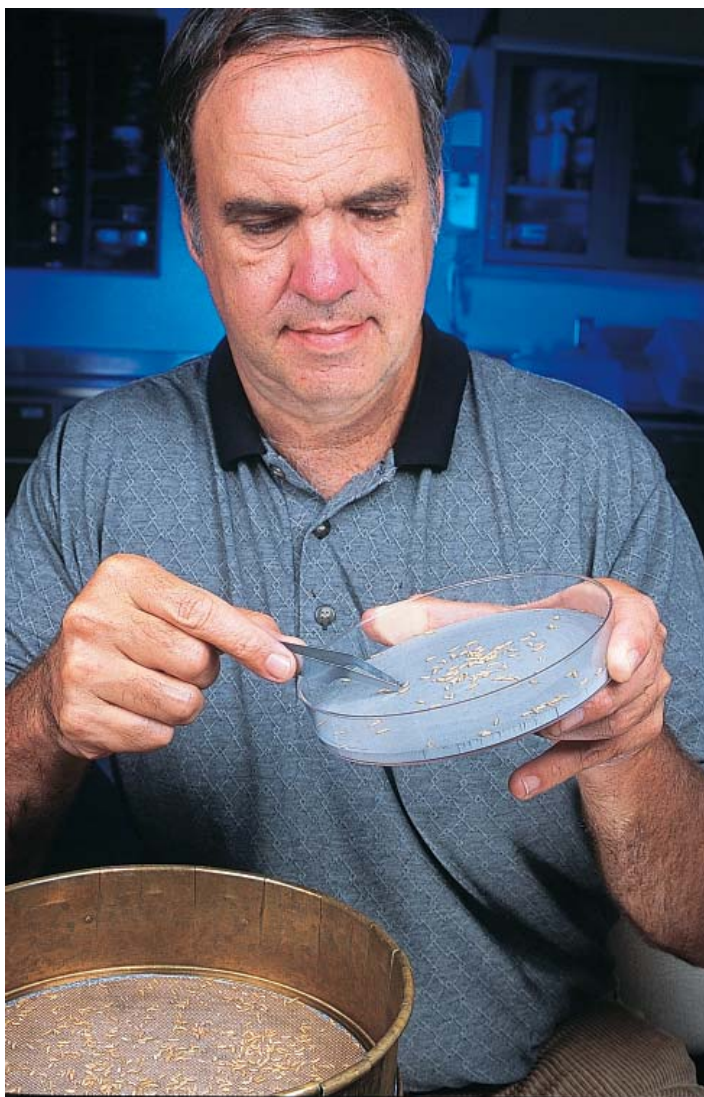
Traps allow warehouse and food-processing managers to make better management decisions about the timing and targeting of control practices. These controls, which include sanitation and crack-and-crevice sprays, are more cost-effective and have less environmental impact than widespread use of conventional chemical treatments.

Use of insect-resistant packaging combined with effective monitoring and the prudent use of pesticides will ensure that consumers receive the highest quality and safest food products possible.—By **Linda McGraw**, formerly with ARS.

This research is part of Crop Protection and Quarantine, an ARS National Program (#304) described on the World Wide Web at <http://www.nps.ars.usda.gov>.

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PEGGY GREB (K9722-1)



Entomologist Frank Arthur collects red flour beetle larvae for testing.

PEGGY GREB (K9723-1)



Left: Red flour beetle larvae exposed to concrete treated with the insect growth regulator hydroprene failed to grow to adulthood. Right: Healthy adults on untreated concrete.

Bioenergy

Today

Taking the lead for the nation last year, the U.S. Department of Agriculture ordered all its locations nationwide to run vehicles and equipment on ethanol or biodiesel blends wherever feasible. Gasoline-operated vehicles and equipment will use a blend of at least 10 percent ethanol and 90 percent conventional gasoline. Diesel vehicles and equipment will use B20 biodiesel, a blend of 20 percent biodiesel and 80 percent regular diesel. The USDA fleet includes more than 700 “flex-fuel” vehicles, which use a blend of 85 percent ethanol and 15 percent gasoline.

The Department’s action reflects the

KEITH WELLER (K9835-17)



At the Beltsville Agricultural Research Center, quality assurance specialist David Johnson examines a sample of biodiesel while Greg Meyer, driver from a cooperating fuel company, fills a 20,000-gallon tank at a boiler plant, which heats BARC’s dairy buildings.

federal government’s commitment to expanding its use of biofuels and biobased products to set an example for the private sector. The year 2000 also saw a groundbreaking for a \$20 million National Ethanol Research Pilot Plant in Edwardsville, Illinois. When completed in January 2003, this will be the largest ethanol pilot plant in the country. The Agricultural Research Service is administering the federal government’s contribution of \$14 million toward the \$20 million cost of construction. Industry considers the plant essential to meeting its goal of increasing annual ethanol production to 16 billion gallons over the next 10 to 15 years.

The new plant is designed to enable researchers to develop the technologies required to improve the efficiency of ethanol production. The more efficient the production, the lower the costs—in terms of both energy and money—and the more competitive ethanol becomes. Since each crop has its own mix of complex sugars and starches, each requires its own techniques to be economically processed into ethanol. The challenge is to design methods that allow different crops to be processed in the same facility.

Throughout the United States and in several other nations, scores of scientists are researching a wide variety of ways to improve ethanol production. These researchers include those in ARS’ national Bioenergy and Energy Alternatives research program. At two of ARS’ regional research centers—in Illinois and Pennsylvania—their goals are to improve the conversion of agricultural plant materials into ethanol and valuable coproducts, lower production costs and fuel emissions, and enhance performance properties of biodiesel. The Western Regional Research

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enhance the
performance of
biofuels.*



KEITH WELLER (K9842-10).

KEITH WELLER (K9842-2)



Above: In a biodiesel-powered tractor, animal caretaker Angel Santiago heads to a dairy barn at BARC. The center uses B20, a common biodiesel blend, in its entire fleet of over 150 diesel vehicles.

Left: High-voltage electrician Alvin Coates fills the tank of an aerial hydraulic lift bucket truck with soy-based diesel.

Biofuel *Basics*

Biodiesel is a clean-burning alternative fuel that can be made from materials such as vegetable oils, animal fats, and spent cooking greases. Typically, biodiesel is prepared by the reaction of fat or oil with alcohol under alkaline conditions. Soy-based biodiesel is the most commonly used form.

Ethanol is an alcohol-based fuel produced by fermenting sugars from crop starches. Currently, 95 percent of ethanol is produced from corn kernels. About 5 percent of U.S. ethanol is made from sugar- and starch-containing materials other than corn. These include wheat, barley, and sorghum grains; sugarcane; cheese whey; and wastes from paper mills, potato processing plants, breweries, and beverage manufacturers—or some combination of these materials.

Originally, most ethanol was made through **wet-milling**, which means the starch is separated from the corn germ and fiber and liquefied by cooking. The liquefying creates sugars in a form that can be fermented with yeast to produce ethanol and carbon dioxide. The ethanol is then removed from the slurry.

The number of ethanol plants has surged in the past few years, and **dry-milling** is now the method used for over half of the ethanol currently produced. In this process, kernels are ground to a fine powder, and all of it is cooked to liquefy it, without removing the germ or fiber. Different enzymes are added at different stages and temperatures as the mash cools, producing ethanol and carbon dioxide.—By **Don Comis**, ARS.

Center, in California, focuses its biofuel efforts on ethanol. In 1999 the national program was expanded to include the breeding of improved energy crops.

Seeing Is Believing

USDA alone last year used over 100,000 gallons of biodiesel fuel and expects to easily double that amount next year. This puts practical examples of biofuel use within driving distance of

fleets to adopt biodiesel, particularly in Maryland. The cities of Greenbelt, Takoma Park, and Ocean City have all recently adopted biodiesel fuel for their snowplows and other public-works vehicles and equipment. Greenbelt also uses biodiesel to run its Connector bus, which ferries Greenbelters on short trips within the city, filling gaps in the Washington, D.C., metropolitan area public transit system. These cities

corn kernels for ethanol, such as *Amaizing Oil*, a new corn oil that can lower blood cholesterol levels, and a valuable food ingredient called *Zeagen*, a corn fiber gum. Both products were found in the fibrous hull that forms the kernel's outermost layer, and both are moving closer to the marketplace.

ERRC engineers have developed a radical alternative way to produce ethanol at a price expected to be significantly lower than is typical of conventional methods. It's called continuous fermentation with stripping. The method removes, or strips, ethanol contained in the escaping carbon dioxide, which is then recycled back to the fermentation vat. In the conventional process, when the ethanol level rises too high in fermentation vats, it slows the yeast's ability to produce more ethanol. The new method continuously strips ethanol from the fermentation broth, freeing the yeast to make additional ethanol.

The team has also developed a new process called pervaporation, which uses a membrane to filter ethanol out of the broth.

Says engineer Frank Taylor, "We are now looking for companies interested in taking our processes off the research bench and develop them further for commercial use."

Andy McAloon is the team's cost engineer. He developed a computer model that can estimate the cost per gallon of ethanol if a new process were used to produce it. "This guides our research so that we don't spend too much time on processes that would not yield a more competitive product," says Kevin Hicks, who leads the ERRC ethanol team. "It's expensive to test a process at the pilot-plant stage, so this model could screen out processes not likely to be practical."

Other researchers at ERRC are working to reduce the cost of biodiesel production. They are making biodiesel fuel from lower quality starting materials, such as soybean soapstock (see story on page 9).

KEITH WELLER (K7776-1)



To help lower the cost of ethanol production, ARS scientists have developed valuable coproducts from corn, such as *Amaizing Oil*, which can lower blood cholesterol levels. Here, chemist Kevin Hicks checks the color and quality of a corn fiber oil sample.

public and private organizations around the country.

Leading by example, USDA's Henry A. Wallace Beltsville (Maryland) Agricultural Research Center (BARC) uses B20 biodiesel in its entire fleet of 150 diesel vehicles, including a tour bus (see cover photo) for the ARS National Visitor Center, located on BARC grounds. The center also heats some buildings with B5 (5 percent biodiesel).

The success with biodiesel in vehicles has encouraged commercial and public

learned about biodiesel by sending representatives to BARC meetings.

Cheaper Ways To Make Ethanol

Scientists at ARS' Eastern Regional Research Center (ERRC), in Wyndmoor, Pennsylvania, are working on lowering ethanol's price per gallon on two fronts: developing coproducts to defray costs and lower-cost production techniques and materials.

They've developed a growing number of valuable coproducts of processing

Enzymes for Greater Efficiency

At ARS' Western Regional Research Center, in Albany, California, scientists are creating better enzymes that produce ethanol in a more cost-effective manner.

"About 10 to 15 percent of the energy required to make ethanol goes toward providing the heat to cook the starch," says chemical engineer George Robertson. "The more energy it takes to make ethanol, the less useful it is as a fuel alternative. So we're working on enzymes that can digest the starch and make ethanol production more efficient. That could open up the ethanol market to other grains, like wheat," Robertson says.

To construct these enzymes, the team uses a technique developed in the pharmaceutical industry called directed evolution. Using biotechnology, they take apart key plant genes and re-construct them, introducing mutations.

The mutant genes are then inserted into yeast organisms, where they begin to make, or express, starch-digesting enzymes. The scientists then screen the yeast colonies for their enzyme-producing abilities and select the best ones for another cycle of gene mutation and selection.

"We can do various things to direct the evolutionary path, speeding up development of enzymes with desired characteristics," says chemist Dominic Wong.

In the laboratory, at 98.6°F, their high-powered enzymes break down starch 50 times faster than the original enzymes. And the technique shows promise of making even better enzymes. The team plans to use similar approaches to develop new enzymes for use in biomass conversion.

Microbes, Too, Can Play a Role

Making biofuels, such as ethanol, economically from the whole crop instead of just the grain is the long-range goal of scientists in the Fermentation Biochemistry Research Unit at ARS' National

Center for Agricultural Utilization Research, in Peoria, Illinois.

"But our starting point is researching fermentation of fiber in just the corn kernel," says ARS microbiologist Rodney J. Bothast, who leads the project. Currently, the kernel fiber is separated out and used as inexpensive cattle feed that is valued for protein, not fiber. If technology were developed to break down the different polymers in kernel

component is not fermented but can be burned to produce energy.

"So far, the most effective way we've found to break down the fiber is to pretreat it with mild acid and then with alkaline hydrogen peroxide," says Bothast.

The pretreated fiber contains sugars, mainly arabinose and xylose and some glucose. Normally, ethanol-producing microbes eat the glucose first, leaving

KEITH WELLER (K7408-6)



Microbiologist Rodney Bothast (left) and technician Loren Iten add starter microorganisms to pilot-plant-size bioreactors in which ethanol is brewed from sugar mixtures derived from corn fiber.

fiber to simple sugars, about 10 percent more ethanol could be produced from each bushel of wet-milled corn.

Bothast collaborates with scientists at ERRC and in the Department of Wood Science at the University of British Columbia, Vancouver, in research on the physical and chemical pretreatment of fiber. Pretreatment frees the cellulose from hemicellulose, starch, and lignin components.

Cellulose fragments are more readily converted into sugars that can be fermented to make ethanol. The lignin

little appetite for the other sugars. Nancy N. Nichols, microbiologist, and Bruce S. Dien, chemical engineer, have developed genetically engineered microorganisms that consume the sugars at nearly equal rates.

These researchers are collaborating with others at the University of British Columbia, Purdue University, and Williams Energy Service, in Pekin, Illinois—the second largest ethanol producer in the country—to test these new microbes on kernel fiber converted to sugars by industrial processes.



Scientists are creating enzymes that produce ethanol in a more cost-effective manner. Technician Tina Williams and chemist Charles Lee use an automated liquid handler and a microplate reader to measure enzyme activity.



At WRRC, technician Sarah Batt uses a robot to pick yeast colonies and transfer them onto starch plates, where they'll be screened for desirable enzyme production.

Value-Added Products

As the scientists seek ways to increase ethanol production efficiency, they're mindful of coproducts that might help make ethanol crops more economically successful. For example, other microbes developed by Nichols and Dien convert the sugars derived from kernel fiber into lactic acid. Biobased companies use lactic acid to produce solvents and biodegradable plastics.

Badal Saha, an ARS chemist, and microbiologist Timothy Leathers have developed yeasts that convert the xylose derived from corn fiber into xylitol, a low-calorie sweetener. Xylitol, which has a minty-cool taste, is used in some mints and gum and sells for about \$3 per pound. It's made from birch wood by an expensive, energy-intensive process.

Saha and Leathers have also discovered fungi that produce enzymes especially well suited for converting corn fiber into sugars. Use of enzymes decreases the amount of acid needed to convert corn fiber to sugars, and that makes ethanol an even more environmentally friendly fuel.

And Out on the Range . . .

Instead of making ethanol from the sugars and starches in plants, Ken Vogel, with ARS in Lincoln, Nebraska, is experimenting with using cellulose and hemicellulose from switchgrass as another source of ethanol. Vogel's hope is that farmers might be able to grow this native prairie grass on highly erodible soils—including those set aside for USDA's Conservation Reserve Program—harvest the grass periodically for ethanol production, and reap conservation benefits, such as reduced soil erosion and enhanced carbon storage.

Vogel and colleagues are breeding new switchgrasses for biofuel use. They're genetically improving the grass for conversion to ethanol and conducting on-farm trials to obtain economic information on production costs. Ron Follett, at Fort Collins, Colorado, is working

with Vogel and ARS scientists at Mandan, North Dakota, to study carbon storage on lands grown for biofuel crops.

Plant geneticist JoAnn Lamb and colleagues at the ARS Plant Science Research Unit in St. Paul, Minnesota, are looking at alfalfa as another cellulose source for producing ethanol. They received \$288,000 from ARS' new \$2.4 million in funding for developing bioenergy crops.

They are breeding a new alfalfa variety specifically to double as a high-quality livestock feed and a bioenergy crop. They'll incorporate genes from southern European varieties to give the plant a thicker, almost woody, stem. This means more cellulose for ethanol production.

The humid East might prefer alfalfa to switchgrass as an ethanol source, but switchgrass is ideally suited for the arid West, because it needs very little rainfall to grow. Both alfalfa and switchgrass can also be burned to generate electricity.

There are obstacles to overcome when making ethanol from cellulose in plants like switchgrass or alfalfa, such as finding ways to convert the complex sugars in cellulose into simple ones that can be fermented to produce ethanol. Facilities to do this conversion will have to be built. Equipment for this purpose could be tested at the new Illinois pilot ethanol plant when it is up and running, as could equipment for the new continuous fermentation stripping process.—By **Don Comis**, ARS. **Ben Hardin** and **Kathryn Barry Stelljes**, both formerly with ARS, also contributed to this story.

This research is part of Quality and Utilization of Agricultural Products (#306) and Bioenergy and Energy Alternatives (#307), two ARS National Programs described on the World Wide Web at <http://www.nps.ars.usda.gov>.

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Zeroing In on a Confectionery Sunflower Blemish

Consumers naturally prefer that their confectionery sunflower snacks look good as well as taste good. So a couple of years ago, when sunflower farmers began seeing mysterious brown spots on the blunt end of their seeds, Agricultural Research Service scientists answered a call for help. Were the spots caused by a disease? An insect? Or both?

An answer and a solution to the problem remain crucial because farmers who produce sunflower seeds for the confection market end up selling the seeds for birdseed at low prices if more than 0.5 percent have the condition called kernel brown spot. Last year, the spots were found in 7 percent of seed samples from some fields.

“At first, we considered the type of fungus called *Alternaria* as a prime suspect,” says ARS plant pathologist Thomas J. Gulya, of the Red River Valley Agricultural Research Center in Fargo, North Dakota.

But Gulya and ARS entomologist Laurence D. Charlet exonerated the fungus and are now pointing at the lygus bug, also known as the tarnished plant bug. It’s an insect whose notoriety has been associated largely with cotton, but

hundreds of crops serve as hosts. In the Northern Great Plains, the quarter-inch-long lygus bugs thrive on increasing acreages of canola. Other tasty crops in the region include sugarbeets, safflower, buckwheat, and crambe.

Scientists at North Dakota State University are working with the ARS scientists to compare lygus bug populations in sunflower fields planted next to certain other crops. “So far, we’ve observed that sunflower, being a late-seeded or late-maturing crop, serves as a host plant for second-generation lygus bugs,” says Charlet.

Though lygus bugs don’t eat much—probably not enough to reduce sunflower crop yields—they have the nasty habit of injecting plant tissues, such as the developing seeds, with digestive enzymes and extracting nutrients with their pointy little mouthparts. The microscopic injuries thwart development of surrounding tissue and appear as big brown spots after the seed matures and is marketed and hulled.

In greenhouse studies, the scientists found kernel brown spot only in seeds from flowers they had covered with bags containing lygus bugs. In USDA insecticide trials at four sites, researchers

found less severe kernel brown spot where the plants had been sprayed well before harvest time with insecticides used to control the banded sunflower moth and the seed weevil. The same insecticides kill lygus bugs. To start learning what steps to take and when best to take them to minimize kernel brown spot, the scientists set up several types of experiments last summer.

The confectionery sunflower market has grown rapidly in recent years to sales in multimillions of dollars involving China alone. U.S. exports of the edible seeds to China grew from 300 metric tons in 1995 to 10,000 tons in 1999.—By **Ben Hardin**, formerly with ARS.

This research is part of Crop Protection and Quarantine, an ARS National Program (#304) described on the World Wide Web at <http://www.nps.ars.usda.gov>.

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EDWARD MCCAIN (K4878-14)



Keeping Cattle Cool Makes *Sense*

During hot summer days, Americans go swimming, drink lemonade, or move inside to air conditioning to cool down. But cattle stay outside, with nothing to cool them. This reduces their growth and efficiency, ARS scientists say.

While nobody is suggesting placing cattle on beach blankets, with sunglasses and strawberry daiquiris to sip, researchers at the Roman L. Hruska U.S. Meat Animal Research Center in Clay Center, Nebraska, have new information to help producers recognize when their cattle are stressed by heat and humidity, which should aid in planning for relief when a heat wave occurs. A heat wave is 3 or more consecutive days of extremely hot conditions.

Hot summers have always affected farmers in certain areas of the country. Heat waves, though, occurred more often in the 1990s than in the previous four decades. Some also lasted longer and were more intense than those of the past, according to recently retired agricultural engineer and biometeorologist G. LeRoy Hahn.

Heat waves are usually most severe from mid-June to mid-August, when many cattle are near market weight. In the heat wave of 1997 farmers lost \$28 million and in the one of 1999, \$40 million, because of cattle deaths and decreased performance. During heat waves, beef cattle do not grow as fast or as efficiently, and dairy cattle don't produce as much milk. Production goes down even more when temperatures remain too warm at night for the animal to recover from the day's heat.

There are a few ways to observe whether cattle might be heat-stressed. A simple way is to compare the temperature and humidity to a graph to see whether the animal is in the danger area. But Hahn thinks that "the animal is the best sensor," and

respiration rate is a way to measure heat stress. When humans get hot, they sweat. Cattle, on the other hand, do little sweating. They lose heat mainly through respiration and, eventually, panting.

During hot days, Hahn says, farmers should count the breaths per minute of a few cattle to see if they exceed the healthy rate of 60 to 80. This can be done with a simple stopwatch, but colleague Roger Eigenberg, an agricultural engineer, has developed a respiration monitor that can be attached securely to the cow to record its breathing rate. This device is currently available only as a research tool.

There are two general ways to help the animal when it's heat-stressed. One way is with a sprinkler system. Agricultural engineer John Nienaber notes that watering should not be done constantly; the animal needs some time to dry before you wet it again because evaporation is what keeps it cool. Also, continued watering creates muddy conditions. Another way is to provide shade or shelter for the animal, but this can be expensive. Hahn thinks the cost of keeping the animal cool is a form of insurance against death losses in extreme heat waves and that it can save money down the road by improving production even during less-stressing conditions.—By **David Elstein**, ARS.

This research is part of Animal Well-Being and Stress Control Systems, an ARS National Program (#105) described on the World Wide Web at <http://www.nps.ars.usda.gov>.

LeRoy Hahn, Roger Eigenberg, and John Nienaber are with the USDA-ARS Roman L. Hruska U.S. Meat Animal Research Center, P.O. Box 166, Clay Center, NE 68933-0166; phone (402) 762-4100, fax (402) 762-4273, e-mail nienaber@email.marc.usda.gov, eigenberg@email.marc.usda.gov, hahn@email.marc.usda.gov. ♦

KEITH WELLER (K4117-15)



Watering cattle and providing shelter are two important ways to help keep them cooler and less stressed during heat waves.

Nematodes: Lords of the Flies?

Biting flies that pester cattle could soon get a taste of their own medicine. Agricultural Research Service (ARS) and University of Nebraska-Lincoln (UN) scientists are testing a way to fight the flies in feedlots where they gather and breed. They're using tiny parasitic roundworms called nematodes that prey on the flies' maggot offspring.

Exploring new, nonchemical ways of protecting cattle is the objective of a 3-year-old project by entomologist David B. Taylor at ARS' Midwestern Insect Livestock Research Unit in Lincoln. In nematodes he sees a biocontrol agent that could be part of an integrated fly-control program at the feedlot along with traps, manure management, sanitation measures, and parasitic wasps.

Since 1999, Taylor and UN associate Thomas Powers have screened 20 species and 50 strains of fly-infecting nematodes for their abilities. Of particular interest were those capable of persisting in cow manure long enough to kill house and stable fly larvae over an entire season.

In Nebraska, where beef cattle are the top agricultural commodity with annual sales of \$5.5 billion, stable flies are considered even worse pests than house flies. That's because attacks by swarms of these relentless biting flies cause blood loss, stress, and feed-efficiency problems. The flies may also harbor disease organisms, and they cost the U.S. beef and dairy cattle industry up to \$1 billion in annual production losses.

Taylor's and Power's strategy calls for battling the pests in manure around feedlots or in soiled calf pen bedding. That's where 80 percent of the flies' brood hatch and feed. And prolific breeders they are—hundreds of maggot offspring emerge from a single pound of manure. Therein lies the nematodes' appeal, for a mating pair of these roundworms can produce 5,000 to 10,000 offspring in a single maggot in less than 2 weeks. Says Taylor, "The nematodes actually reproduce faster than the flies."

In experiments, up to 99 percent of fly maggots died within 48 hours of infection by the top fly fighter, *Steinernema feltiae*. In the laboratory, "the nematodes can live in bovine manure for 4 to 6 weeks without hosts," says Taylor. In feedlots, he adds, "We'd like to apply them in May and get season-long fly control." Chemical insecticides, in contrast, must be reapplied, and flies can develop resistance to them.

The researchers are testing ways to apply the nematodes on manure and protect them from drying and ultraviolet light. About 1 million nematodes per square meter are used, but lower rates might suffice, keeping the costs closer to chemical controls. If the approach works, the nematodes could also be used to fight corn rootworms in manure-fertilized fields, says Taylor.—By **Jan Suszkiw**, ARS.

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Electrical Conductivity Spots Salty Soils

Electrical conductivity (EC) measures the amount of salt in a field as well as the field's composition—the amount of sand, clay, and organic matter. Farmers want to know the composition of their soil so that they can apply the correct number of seeds and chemicals to each section of their land. A farm with varying soil composition can be subdivided into sections according to EC data.

ARS agricultural engineer Hamid Farahani and his fellow researchers at the Water Management Laboratory in Fort Collins, Colorado, have shown that EC data can be a practical tool in determining how effective changes in irrigation water management practices have been in minimizing the buildup of salts in the crop root zone. In cases where there is no buildup of salts, any measured variability in EC would reflect changes in soil composition across the field.



Farahani uses a pickup truck to pull a machine that measures electrical conductivity of the soil. As the truck maneuvers over the field, two EC readings are taken every second: one that measures the top foot of the soil and another that measures the top three feet. A 140-acre field can be driven in 6 hours, giving about 14,000 data points.

A Global Positioning System (GPS) mounted on the truck links to satellites and tells a computer exactly where each data point is in the given field. This is similar to the devices found in some cars that can locate them if stolen.

Farahani puts the information collected into a special computer program to get an indication of the changes in salt loads across the fields. Different colors show the amount of EC within the field. With this machine, farmers can quickly get a map of their field's variability. Using the EC map as a guide, farmers only need to collect a few soil samples from each specific EC area to determine soil composition and decide whether or not to modify management.

Without this machine, it would take days to collect enough samples to make a similar map, and it would cost significantly more.

Farahani has gone back to survey farms 2 or 3 years later to find only small changes in EC, which indicates good irrigation practices. Drastic changes would have indicated problems in the overall management of the irrigation water.—By **David Elstein**, ARS.

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New Rice Could Benefit Malnourished Populations

SCOTT BAUER (K9950-1)



Geneticist J. Neil Rutger compares golden-hulled low-phytate rice (left) with rice that lacks the color-coding gene.

In ancient Asian civilizations, rice was believed to be a gift from the gods. Its influence on various cultures is still evident. Rice is revered today for its nutritional value and plays an important role in the diets and economies of nations around the world.

And now, rice grains that contain less phytic acid could mean better nutrition for the world's malnourished peoples, more nutritious animal feed, and less potential for water pollution from manure.

Humans need minerals to stay healthy, and people rarely have phosphorus deficiencies. But cereals like rice store most phosphorus in the grain as phytic acid, which can't be digested by one-stomached animals like fish, chickens, pigs, and humans. It binds to minerals such as iron, calcium, magnesium, and zinc in the slightly acidic conditions in our intestines. Because phytic acid is poorly digested and used, the minerals it binds to are less available to our bodies.

While phytic acid is involved in many necessary roles in seeds, people in nations with mainly grain-based diets could use less of this compound in their food. Livestock too could still be healthy with less of it in their feed. And the environment would benefit if less undigested phosphorus were excreted in manure, because it can lead to pollution of lakes and streams.

Scientists at the Dale Bumpers National Rice Center in Stuttgart, Arkansas, wanted to reduce the amount of phytic acid in rice. J. Neil Rutger, director and supervisory geneticist at the center, produced new breeding stock—or germplasm—for creating improved varieties. He enlisted the expert assistance of geneticist Victor Raboy, who developed the patented technique that yields grains with lower amounts of phytic acid. Raboy, based at the ARS' Small Grains and Potato Germplasm Research Unit in Aberdeen, Idaho, used the technology to develop new types of corn, barley, and soybeans. This is the

first time his technique has been used to produce low-phytic-acid rice. The resulting rice contains only half the phytic acid of its parent, which translates to enhanced nutritional value.

For more than 30 years, Rutger has worked with rice breeding lines to enhance desirable qualities and diminish weaknesses. To create a new, low-phytic-acid plant, Rutger selected Kaybonnet—one of the most popular types of rice grown in Arkansas—when the project began in 1994. Germplasm he developed was sent to Raboy's laboratory, where it was screened for low-phytic-acid mutations. The first such mutation was

SCOTT BAUER (K9952-1)



Bottom: Parent variety of rice with normal phytic acid. Middle: The new low-phytic-acid variety. Top: The new variety given a gene for golden hull color to help identify it.

dubbed KBNT *lpa1-1* (short for Kaybonnet low-phytic-acid gene 1). Initial analyses showed that phytic acid was reduced by 45 percent in this rice. Later, Steven R. Larson, then a postdoctoral scientist with Raboy and now with ARS' Forage and Range Research Laboratory in Logan, Utah, carried out the genetic mapping of KBNT

lpa1-1 using populations Rutger constructed for this purpose.

To further study the phytic acid content of this new breeding line, chemist Rolfe J. Bryant at Stuttgart and Rutger compared it to common cultivars and to its parent. First, different lines were milled. Milling, often called whitening, removes the outer bran layer of the rice grain, leaving a core that is mostly carbohydrates. Milling makes brown rice become white rice. Vital nutrients are found in the bran, including about two-thirds of the phytic acid.

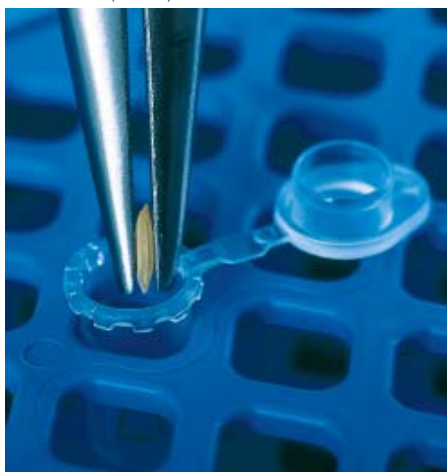
Rutger worked with Bryant to analyze the new line and other varieties. Bryant compared samples from different degrees of milling and examined their phytic acid content. He found that the total phytic acid concentration in the brown rice of KBNT *lpa1-1* (before the bran is removed) was 49 percent lower than that of its parent—a characteristic improved through breeding. KBNT *lpa1-1*'s phytic acid content before milling was also 25 to 52 percent less than other varieties they tested, with an average of 42 percent less. Although a small amount of phytic acid appears to remain in the white rice even after intensive milling, the phytic acid content of KBNT *lpa1-1* was still less than or equal to that of its parent.

Although there is less phytic acid in the new line, it has more phosphorus available for digestion and absorption by the body. One trade-off was a 10-percent lower grain yield than its parent. But decreased yield is common initially with many crop breeds and could be bred out eventually, Rutger says.

"If used in animal feeds, the bran portion of KBNT *lpa1-1* rice should be of greater nutritional value than brans from other rice varieties," Rutger points out. "This means less undigested phosphorus in the animal's manure."

ARS and the University of Arkansas released the new rice to breeders and researchers earlier this year. Rutger and Raboy are continuing to screen for other genes with the low-phytic-acid traits.

SCOTT BAUER (K9954-1)



A rice kernel is crushed with pliers and inserted into a microcentrifuge tube for phytic acid extraction and analysis.

The next phase of their research was to make low-phytic-acid rice visually distinguishable from other varieties now in the marketplace. To do that, the scientists borrowed a color-imparting gene from a golden-hulled rice no longer marketed and introduced it into KBNT *lpa1-1*. Then they selected resulting lines that had both low-phytic-acid characteristics

SCOTT BAUER (K9953-1)



Chemist Rolfe Bryant mixes a rice extract with a reagent that will reveal the amount of phytic acid in the kernel.

and a gold hull. The golden color of new rice will differentiate it from other varieties and prevent mixups.

Rutger hopes studies will be held where volunteers will eat meals prepared with the new rice to see whether their mineral absorption increases. That's what happened to volunteers who participated in a study of low-phytic-acid corn that Raboy developed. (See "Feeling Weak? Try the Tortillas!" *Agricultural Research*, March 2000, p. 13.)

"Volunteers who ate tortillas made with low-phytic-acid corn flour absorbed 50 percent more iron than those who ate tortillas prepared with conventional corn flour. We expect similar results with volunteers who eat KBNT *lpa1-1* rice," Raboy says.

"This rice line would be of significant nutritional value to developing nations where mineral deficiency is common," Rutger says. "There has been a great deal of interest in our research from organizations in various nations."—By **Jim Core, ARS.**

This research is part of Plant, Microbial, and Insect Genetic Resources, Genomics, and Genetic Improvement, an ARS National Program (#301) described on the World Wide Web at <http://www.nps.ars.usda.gov>.

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Worldwide Effort To Map the **Bovine Genome**

KEITH WELLER (K9977-1)



Molecular biology technician Renee Godtel prepares bovine DNA samples for sequencing.

First there was the mapping of the human genome. Then, this spring, scientists announced they had nearly completed the genome mapping of the mouse. Now, scientists are in the early stages of mapping the bovine genome to help produce cattle with improved production traits and to possibly help in finding cures for human diseases.

The Agricultural Research Service is part of a group of government and university laboratories from four continents in the initial stages of mapping the bovine genome. The ARS research effort is led by Steven Kappes and John Keele of the Roman L. Hruska U.S. Meat Animal Research Center, in Clay Center, Nebraska. Kappes is center director, and Keele is an animal scientist.

The project began in spring 2000, when Kappes started contacting labs from around the world to develop a physical (bacterial artificial chromosome—BAC) map of cattle. Bacteria—and more specifically, bacterial chromosomes—are used as hosts for pieces of bovine chromosomes. The bacterial hosts are used to generate many identical copies of a piece, or clone, of cattle DNA. The BAC map will be a useful tool for identifying genes that affect production traits in farm animals and an excellent resource to improve the efficiency of a future effort to sequence the entire bovine genome.

The first step in the process is to fingerprint individual BAC clones. Researchers at the British Columbia Cancer Agency Genome Sciences Centre have been funded to construct a fingerprinted BAC map. A fingerprint is obtained by cutting DNA from a BAC clone into pieces and separating the fragments on a gel. The fingerprint pattern of the different fragments is used to identify overlapping BAC clones. A BAC map is the collection of overlapping clones that represent the entire bovine genome.

Funding for this part of the project has been provided by USDA-ARS; the Biotechnology and Biological Sciences Research Council and Roslin Institute, from the United Kingdom; the University of Alberta and the Alberta Science and Research Authority, in Canada; and the University of Illinois.

The fingerprinting will be performed on 280,000 BAC clones from two libraries constructed by scientists at the Children's Hospital Oakland (California) Research Institute. The first BAC library was constructed from Holstein bull DNA and the second from Hereford bull DNA. Each BAC clone contains about 170,000 bases of cattle DNA.

The second step, which can occur simultaneously with fingerprinting, is sequencing both ends of all 280,000 clones. This work is being conducted by ARS, the University of Illinois, Texas A&M University, AgResearch of New Zealand, the Commonwealth Scientific and Industrial Research Organization of Australia, the Brazilian Agricultural Research Corporation, and the University of Alberta. The Institute of Genomic Research in Rockville, Maryland, has been contracted to do some of the sequencing. The National Institute for Agricultural Research in France is fingerprinting and end-sequencing clones

from a BAC library constructed in their laboratory. They will combine their information later with the international effort. Kappes is talking with other organizations to help with the end sequencing.

The scientists will combine the end-sequencing and fingerprinting information to determine the overlapping BAC clones. Kappes says, "Ideally, we would like one set of contiguous overlapping clones—'contigs'—for each of the 30 chromosomes in the bovine genome. But it's likely that we'll have gaps between several contigs for each chromosome."

So far, 249,000 of the 280,000 cattle BAC clones have been fingerprinted, and the end-sequencing effort is under way. The completion date for the bovine BAC map is February 2003.

The researchers hope that the next phase of the project will be sequencing the bovine genome. Kappes and other scientists have sent a proposal to the National Institutes of Health (NIH) to do this work.

The BAC map alone costs \$4.5 million, while NIH estimates it may cost \$100 million to sequence the bovine genome to a finished stage. Kappes says the effort is expensive, but it will have many tangible benefits. Scientists from ARS and elsewhere will use the BAC map and sequence information to improve productivity traits in cattle. This means they may be able to more accurately select genetically superior animals for specific needs, such as lean beef, milk production, reduced feed requirements, and improved health and welfare. This ability would increase the profitability of beef production. The research should also benefit those who raise sheep, since the genetic makeup of sheep is very similar to that of cattle.

This research may also help the medical community. "As we define certain biological mechanisms in livestock, the information may benefit human medicine," says Kappes. "We are currently defining a genetic mechanism affecting muscling in sheep. This is of specific interest to research efforts in human medicine because a similar mechanism is observed in cancer cells."

Not only is there similarity in the DNA sequences of genes in farm animals and in humans, but also the biological processes are very similar across species. Eventually, researchers will be able to compare the human genome to the bovine genome to help determine the function of genes for both livestock production and human well-being.—By **David Elstein**, ARS.

This research is part of Food Animal Production, an ARS National Program (#101) described on the World Wide Web at <http://www.nps.ars.usda.gov>.

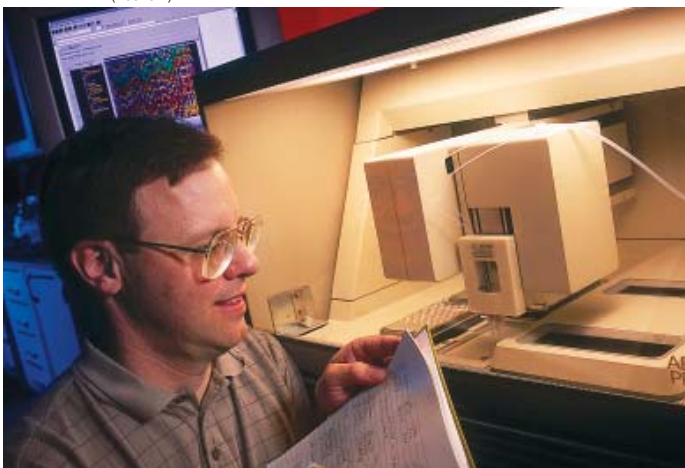
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KEITH WELLER (K9978-1)



Along with the genomics group at MARC, animal scientist John Keele is part of the team that's working to produce a BAC map of the bovine genome.

KEITH WELLER (K9976-1)



Chemist Tim Smith observes an automated DNA sequence instrument, which produces 96 bovine DNA sequences every 3 hours.

DUANE SMAILUS (K9976-20)



At the British Columbia Cancer Agency Genome Sciences Center in Vancouver, Canada, genomics technologist Pawan Pandoh prepares BAC fingerprints used in creating the bovine map.

Leading the Way in **CO₂ Research**

SCOTT BAUER (K10044-1)



While visiting the ARS Photosynthetic Research Unit in Urbana, Illinois, soil scientist Bruce Kimball (right) assists postdoctoral researcher Carl Bernacchi as he uses a portable photosynthesis analyzer on soybean plants.

SCOTT BAUER (K10050-2)



To determine the effect of elevated CO₂ on plant growth, University of Illinois students harvest aboveground growth of soybean plants.

Carbon dioxide. CO₂. Some think of the gas in positive terms, such as making plants grow or trading carbon credits. Others think that its rising levels in the atmosphere may be harming the environment.

The Agricultural Research Service has scientists stationed around the country to study CO₂ and its effects on agricultural systems—particularly, how it will affect plants in the future and how we can capture it more effectively in agriculture. This article provides updates on some of these studies. (See also “Preparing Agriculture for a Changing World,” *Agricultural Research*, July 1997, p. 4.)

Are Western Rangelands the Missing Sink?

Reports from a CO₂-monitoring network across western rangelands show that, at the very least, rangelands seem to be a substantial carbon sink, or storage reservoir, helping slow the buildup of CO₂ in the atmosphere.

The network partially has its roots in a mystery announced by global modelers in the 1980s: They couldn’t account for 2 billion tons of carbon emitted annually in CO₂ from burning fossil fuel, deforestation, and other sources. It had to be stored somewhere on Earth, but modelers didn’t know where. All they knew was that it was stored and that it might help offset possible global warming effects caused by rising CO₂ emissions.

Scientists want to know where this sink is, and how much CO₂ it might be able to hold, so that they can preserve its carbon-storing capabilities and develop expectations for other ways to remove CO₂ from the atmosphere.

Many looking for this missing sink turned their attention to the United States,

especially the Northeast and Great Lakes regions, where newly emerging forests on former farmland might hold the missing carbon in tree trunks, limbs, and roots. But in 1995, ARS rangeland scientists launched a CO₂-monitoring network in the central and western United States to assess rangeland functions, since rangelands cover about half of Earth’s land surface.

Of the 11 sites in the network, the first 3 to report were those in Mandan, North Dakota; Woodward, Oklahoma; and Temple, Texas. Together, the three sites give a north-south cross-section of the Great Plains.

Scientists found that grasslands were actually substantial contributors to carbon storage. Soil in ecosystems represented by the Mandan site was storing 1 million metric tons of carbon each year; the Woodward site, 2 million; and the Temple site, 6 million. All eight other sites had similar findings.

The effects of grazing and fire on carbon storage are now under study at many of the sites. Albert B. Frank, an ARS plant physiologist at Mandan, says he has found that properly grazed lands can store as much carbon as lands that are never grazed. But overgrazing can cause loss of the carbon stored in soil. Woodward researchers are working to find the rate of grazing that optimizes animal production and rangeland health.

Each of the 11 monitoring sites has at least one 6- to 8-foot tower to measure CO₂ exchange among plants, soil, and atmosphere. Each tower has a CO₂ Bowen ratio station—a type of weather station equipped with instruments that take more than 30 measurements, including CO₂ and water vapor gradients. The samplers take readings every 2 seconds and calculate an average CO₂

gradient every 20 minutes, around the clock. Four of the sites also have closed, 1-meter-square chambers for comparing CO₂ uptake at the soil level with information from the CO₂ station.

The network represents the ecosystems that make up America's western rangelands, as well as many ecosystems around the world. The other sites are at Burns, Oregon; Logan, Utah; Dubois, Idaho; Miles City, Montana; Cheyenne, Wyoming; Fort Collins, Colorado; Tucson, Arizona; and Las Cruces, New Mexico. Boise, Idaho, entered the network in mid 2002.

About FACE

In addition to monitoring the exchange of CO₂ between the atmosphere and the land, ARS and other scientists are studying the effects of elevated levels of CO₂ on crop plants.

One such scientist is Donald R. Ort, a plant physiologist who leads ARS' Photosynthetic Research Unit in Urbana, Illinois. In soybean fields not far from Urbana, Ort hopes to harvest a wealth of information from the plants that could foretell how this important legume crop would perform under elevated levels of CO₂ and ozone predicted for the middle of this century, he says.

To do this, Ort and colleagues at ARS and the University of Illinois at Urbana-Champaign (U of I) have encircled portions of the 40-acre field with ringlike devices that fumigate the plants with 550 parts per million (ppm) of CO₂ and up to 150 parts per billion (ppb) of ozone. In the Urbana study, the concentration of CO₂ is about 1.5 times more than the current level of 370 ppm, and ozone levels

are about 1.5 times the current ambient levels of 40-50 ppb in the Midwest.

At ground level, ozone is an air pollutant that is toxic to plants. It is created when photochemical reactions involving gases from industrial and transportation sources occur in the presence of sunlight. When crops such as soybeans are grown in ozone concentrations typical in some regions of the country, photosynthesis

collaboration with about 14 other research teams.

Ort's lab monitors changes in leaf transpiration rates, photosynthetic capacity, and other biological activities of test plants as they're fumigated with CO₂, different concentrations of ozone, or both. All the while, scientists compare the data they glean from the test plants to data from control plants.

Of particular interest are soybean lines whose seed yields seem to rise in response to elevated CO₂. "There are reports that certain soybean cultivars and parentages can take advantage of elevated CO₂," says Ort. "Others may adjust to it by slowing their photosynthetic activity." Use of the fumigation rings also affords an opportunity to explore the prediction that higher CO₂ may help abate ozone damage to plants. One reason may be that the leaf openings, called stomata, constrict at elevated CO₂ levels, so ozone is less able to enter and cause harm at the cellular level.

Also showing their stamina are new snap beans developed by the ARS Air Quality Plant Growth and Development Research Unit, in Raleigh, North Carolina. These beans show high resistance to foliage damage under the heightened ozone conditions of the fumigation rings, whereas sensitive lines show extensive injury. Plant physiologist Kent Burkey is looking for genes associated with the snap beans' ability to use their own vitamin C to defend against ozone.

Next, researchers will begin the search for genetic traits and markers. Bean breeders can one day use these to speed

SCOTT BAUER (K10054-2)



Following ozone exposure, the sensitive snap bean plant on the right is stunted and has some damaged leaves. The plant on the left, however, is more tolerant. ARS scientists are searching for bean genes that can use vitamin C to defend against ozone.

can be suppressed, which lowers yields.

Ground-level concentrations of ozone, like those of CO₂, are increasing. The long-term goal of the 10-year project, called SoyFACE, is to make sure the nation's soybean farmers are well stocked with new varieties of the crop that can tolerate higher ozone, take advantage of the increased CO₂, or both.

"If we wait for global climate change to happen, we're going to be way behind the curve," says Ort. He, along with Stephen Long, a professor at U of I's Department of Crop Science, lead the study—now in its second year—in close

development of new tolerant varieties.

SoyFACE follows earlier Free Air CO₂ Enrichment (FACE) experiments conducted by ARS researcher Bruce A. Kimball and others, near Phoenix. The Arizona group has studied cotton, wheat, and sorghum.

In the late 1980s, researchers from Brookhaven National Laboratory working with Kimball and other ARS and university scientists developed the FACE technology. Now there are more than a dozen FACE projects in the United States and around the world.

Using FACE technology, scientists release large quantities of CO₂ over plots of open-field crops, thereby simulating conditions as representative of future fields as is possible today. Researchers found that crops that use the type of photosynthesis called C₃, such as wheat, rice, and cotton, all increased in yield with elevated levels of CO₂. Cotton's yield increased by 40 percent when CO₂ was added. Wheat yield increased about 15 percent, and the plants used about 7

percent less water. Yields of sorghum, a crop that uses C₄-type photosynthesis, did not increase with an additional 200 ppm of CO₂ at ample water supply, but the plants used about 10 percent less water. On the other hand, under drought conditions, grain yields increased 25 percent on average.

The initial FACE studies were a great success, providing much of the information available on how crops would likely respond to changes in their environment.

Increased Temperatures Have a Chilling Effect

Research at ARS' Crop Genetics and Environmental Research Unit in Gainesville, Florida, indicates that global warming could be more of a problem for seed crops than for forage crops. L. Hartwell Allen, Jr., an ARS soil scientist, says the threat to crops such as cereal grains and legumes isn't rising CO₂, but rather a potential rise in temperatures.

While higher CO₂ levels tend to increase crop yields, elevated temperatures

could leave seed crops sterile. In elevated-temperature studies, either pollination of individual flowers fails completely or, when fertilization is successful, seeds develop poorly. For example, soybean yields are reduced both because fewer seeds are produced and individual grains weigh less.

As a rule of thumb, Allen says, seed productivity decreases about 10 percent for every 2 degrees Fahrenheit the temperature increases above ideal levels. But photosynthesis and plant size are little affected until much higher temperature levels. Even when seed development fails in a plant, it may still grow to its typical size.

Allen, plant physiologist Joseph Vu, and University of Florida crop physiologist Kenneth Boote found that several physiological functions related to reproduction and pollination fail as temperatures increase. Abnormally high temperatures during the short pollination season (2 to 3 weeks) could affect seed development at exactly the wrong time

SCOTT BAUER (K10052-1)



ARS scientists from Urbana and Phoenix join forces in Urbana, Illinois, to measure the effects of elevated ozone on soybean photosynthesis, growth, and development.

SCOTT BAUER (K10049-1)



Plant physiologist Donald Ort monitors output of an ozone generator.

in a plant's life cycle.

Traditional breeding could offer the best hope if, for example, scientists locate wild relatives that grow in very hot environments and incorporate their tolerance into productive crop varieties. Allen says the time of day when pollen is shed in a plant could be a factor in its survival. Varieties that shed pollen earlier in the day, when temperatures are cooler, would be more likely to flourish. Genetic engineering could help if scientists introduce desirable genes from other plants.

Although increased CO₂ levels appear to benefit many plants, it is the interaction of multiple environmental factors that makes predicting the effects of global change on agriculture challenging. Fortunately, agricultural soils also store carbon, where it provides environmental and productivity benefits and helps to alleviate atmospheric CO₂ levels.—By **David Elstein, Don Comis, Jan Suszkiw, and Jim Core, ARS.**

This research is part of Global Change, an ARS National Program

(#204) described on the World Wide Web at <http://www.nps.ars.usda.gov>.

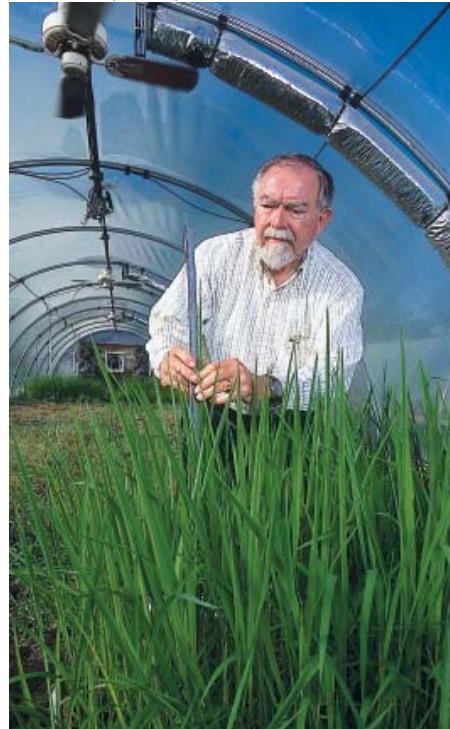
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SCOTT BAUER (K10045-1)



In Gainesville, Florida, soil scientist L. Hartwell Allen measures growth of 1 of 14 rice cultivars grown to determine the effect of elevated temperature on reproductive growth and seed yield.



SCOTT BAUER (K10048-1)



University of Illinois site manager and engineer Tim Meis programs CO₂ and ozone level controls for the distribution station just outside of each CO₂ and ozone FACE ring.

Keeping Tabs on Landscape Changes

Eighty years can be time enough for many changes in a landscape. Or eight decades may be such a short time that there are no discernible changes at all. But how can you tell? How do you track vegetation changes over a period of time that is longer than many lives, let alone most careers?

Rangeland scientists Keith D. Klement and Rod Heitschmidt were able to chronicle 80 years of shifts in vegetation on the Northern Great Plains in a recent ARS publication. They used photographs taken between 1908 and 1937 and reshot about 40 years later. Then, they took a third set of photos of the same locations, giving them a second 40-year interval.

Forty-two sites in Montana, Nebraska, North Dakota, South Dakota, and Wyoming were documented for *Eighty Years of Vegetation and Landscape Changes in the Northern Great Plains. A Photographic Record*. Klement and Heitschmidt, who are with ARS' Fort Keogh Livestock and Range Research Laboratory in Miles City, Montana, planned the publication to give researchers, land managers, naturalists, policymakers, and the general public a way to see even subtle alterations in the Northern Plains over time.

"This series of photographs is important because it's very hard to be completely sure of vegetation and landscape changes, even in places you know well yourself," explains Heitschmidt. "You think you'll remember how a landscape looks. But when you come back a few years later, do you really know how much change has taken place, unless something dramatic has happened?" Written descriptions and plant counts never have the same impact and independent witness that photographs do, he adds.

Homer Shantz, a noted botanist and former president of the University of Arizona, took the earliest set of photographs between July 14, 1908, and September 1, 1937. Shantz began rephotographing the original sites on June 13, 1958, but died later that year. His graduate student, Walter S. Phillips,

completed the repeat photography, shooting until 1960. The third set of black-and-white photographs was commissioned by ARS in 1998. Klement revisited the sites in 1999 to confirm changes in vegetation and landscape.

When they first compared the pictures, Klement and Heitschmidt were most impressed by how few changes in the types of vegetation there had really been, despite the fact that the Northern Great Plains has been ranched extensively during the past 80 years.

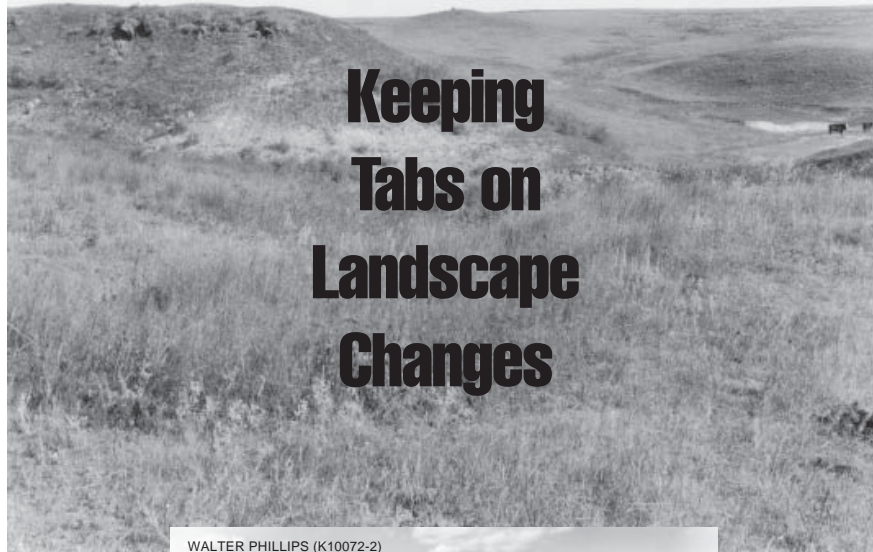
"It wasn't that the land looked better than I expected, because 'better' is a value judgment. We simply examined the photographs for changes in the types and density of plants. That's objective science. And we just didn't see a lot of major alterations," Heitschmidt says.

He points to this as a sign of how stable the Northern Great Plains vegetation complex really is. The plains evolved with tough, perennial grasses to withstand drought and grazing. "Grasses don't care whether the grazing is done by bison or cows, as long as the perennial grasses are left in place and the area isn't overgrazed," he says.

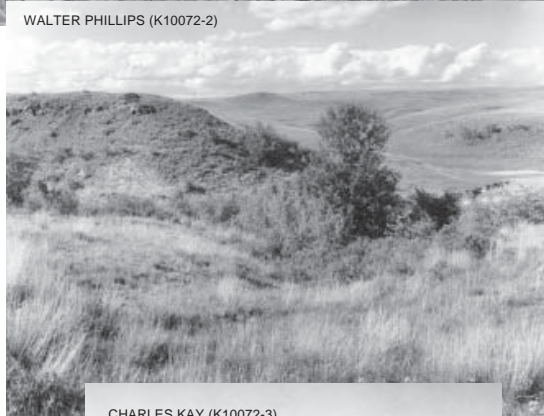
Most marked among the changes the researchers did find were the increased density of ponderosa pine trees on mountains and hillsides and the thickening of sagebrush in valleys and foothills.

"Wildfires have been keeping the pines in check for centuries. But fires have been controlled or eliminated in the last 80 years," Klement says. "Without periodic fires, smaller trees are not controlled. So the natural ecology of larger, older, and less dense ponderosa pine forest with a herbaceous understory has given way to numerous smaller trees, with a barren or pine-needle-filled understory. At many sites, we see a dense forest emerging in place of a historically open type of forest."

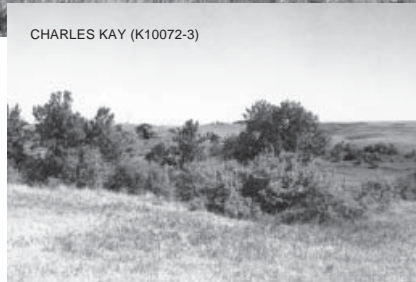
The lack of fires has had a similar effect on sagebrush in the drier lowland areas, that is, an increase in the amount of land covered by sagebrush and the density of each patch.



WALTER PHILLIPS (K10072-2)



CHARLES KAY (K10072-3)



Top to bottom: Photos taken in 1924, 1959, and 1998 at a range about 1.3 miles west-southwest of Fryburg, North Dakota. Since 1924, uplands and hillsides once dominated with grasses now host several shrub and tree species.

Klement also noticed more expansive areas of crested wheatgrass and yellow sweetclover, with the bright yellow flowers of the clover casting color over some grassy stretches. These nonnative species were once planted along roadsides and ditches and introduced into pastures and hayfields. In some cases they are still being planted today. In several areas, they have escaped the planting sites and have begun to spread and outcompete native plants.

Other actions by people have had more subtle effects on the landscape. Fences and roads have created microclimates—new ecological niches. Birds sit on fences and drop seeds where they might not normally fall. Road grading creates raised areas that hold water, which then become hospitable to different plants.

In some photo series, the land can be seen circling back to an earlier state. Photos from 40 years ago show a common practice of clearing vegetation and beaver dams from streams to help the water run more freely for irrigation. “But we’ve learned about the importance and the value of riparian zones now, and in the most recent set of photos, you can see where people have let the plants grow back along the creek banks,” Klement says.

Since the book’s publication, Klement has gotten a steady stream of requests for copies. “All sorts of people have been interested—extension agents, the Bureau of Land Management, universities, historical societies, the Natural Resources Conservation Service, and local ranchers, to name a few,” he says. “State agencies, like the Nebraska Game and Parks Commission, and high school teachers, like one from Maize, Kansas, who plans to use the book as part of his curriculum, are also putting this land record to use.”

Even a woodland ecology scientist from Rockhampton, Australia, plans to compare it to similar work in her country.

“What we have now are preserved reference points that let us clearly see changes that are not apparent within a short span of time,” Klement emphasized. “Hopefully, someone will do this again in another 40 years.”

The publication is available online at <http://www.ars.usda.gov/is/np/eightyyears/eightyyearsintro.htm>.—**By J. Kim Kaplan, ARS.**

This research is part of Rangeland, Pasture, and Forages, an ARS National Program (#205) described on the World Wide Web at <http://www.nps.ars.usda.gov>.

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HOMER SHANTZ (K10072-4)



WALTER PHILLIPS (K10072-5)



CHARLES KAY (K10072-6)



Top to bottom: Photos taken in 1917, 1959, and 1998 at a range in Lewistown, Montana. The cars in the photos change with the era, but each car is positioned next to the same fence post. Between the times of the first two photos, the road was moved back about 100 yards. When comparing the three photos, note the increased density and cover of ponderosa pine on the Judith Mountains in the background.

Chinese Pigs Provide Insight to U.S. Swine Reproduction

Some say the animal is so cute. Others say it is the funniest-looking thing they've ever seen. Either way, scientists in the United States were delighted to obtain some for research in the late 1980s.

They are the Meishan (pronounced MAY-shawn) pigs of southern China. They were always known for their large litters (15-16 piglets compared to 10-12 for U.S. sows). Researchers from various countries wanted to find out why this occurred.

The first Meishan pigs (and a few other Chinese-bred pigs) were brought to the United States in 1989. After being quarantined in Florida, they were delivered to ARS' Roman L. Hruska U.S. Meat Animal Research Center (MARC) in Clay Center, Nebraska; the University of Illinois; and Iowa State University.

Initially, researchers were interested only in the females; male Meishans were brought along simply to help produce more females, says MARC animal physiologist Joe Ford. Scientists hypothesized that elevated levels of follicle-stimulating hormone (FSH) in Meishan females were the cause of their large litters. But the researchers found that FSH levels in Meishan females were similar to those in U.S. females, says Gary Rohrer, a geneticist at MARC.

After further study, they found that Meishan females begin puberty in only 60 days, much earlier than other pigs. They also ovulate more eggs per cycle and have a greater uterine capacity to deliver more live piglets.

Eventually the researchers started studying the males and learned some interesting facts about their reproductive organs, too. The first important finding was that Meishan boars have five to seven times more FSH in their blood than U.S. boars have, but their testes were 40 percent smaller than those of U.S. boars.

The small size reflects the finding that Meishan boars have fewer Sertoli cells—the cells that develop sperm—in their testes. So, fewer Sertoli cells means less sperm and, curiously, more FSH. But how this mechanism relates to litter size or to other reproductive traits is still under study.

ARS researchers also found that a gene (or genes) on the X-chromosome affects the size of Meishans' testes, whereas in rodents, testis size is more affected by the genes on the Y-chromosome.

Scientists like the fact that Meishans provide a new and different model to investigate swine sperm production. Ford says that since 75 percent of newborn pigs in the United States are produced through artificial insemination, finding ways to improve sperm production is important to improving the efficiency of U.S. pork production.

KEITH WELLER (K10089-2)



Geneticist Gary Rohrer examines Meishan pigs at the ARS Roman L. Hruska U.S. Meat Animal Research Center in Clay Center Nebraska.

Pork products from Meishans taste as good as those from conventional breeds, but Meishan meat has more fat. Ford says his laboratory is using the Meishans only as an experimental model to compare to other breeds of pigs. They are not very useful for crossbreeding, but their unusual characteristics help scientists study biological processes in traditional pigs.—By **David Elstein**, ARS.

This research is part of Food Animal Production, an ARS National Program (#101) described on the World Wide Web at <http://www.nps.ars.usda.gov>.

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KEITH WELLER (K10089-4)



Farmers Adopt ARS-Improved Soybean Bacterium

A lowly bacterium first cultured 2 decades ago by ARS researchers is now enjoying celebrity status as a commercial soybean inoculant.

Behind that success is Urbana Laboratories, a St. Joseph, Missouri, company that has sold nearly 14 million acres' worth of the inoculant since obtaining a license from ARS to market the bacterium in 1994.

A *Bradyrhizobium* species, the bacterium converts (fixes) gaseous nitrogen into forms that soybean plants can use for optimal growth and higher yield. In return, the plant shelters and nourishes the bacterium inside root nodules, where nitrogen fixation occurs.

ARS microbiologists L. David Kuykendall and William J. (Jim) Hunter originally developed, tested, and in 1991 patented the bacterium as an improvement over *Bradyrhizobium* strains being used by soybean farmers.

In the laboratory, they used nitrous acid to trigger gene mutations in a liquid culture of *B. japonicum* strain I-110. Through a similar process called direct selection with one of the resulting mutant strains, they settled on a strain called TA11Nod+ (or Nod+) as their final choice.

Interestingly, use of this bacterial genetics approach—rather than a recombinant one—may have contributed to the mutant strain's acceptance among farmers, notes Hunter, with ARS' Plant, Soil and Nutrient Research Unit, Fort Collins, Colorado.

Nod+, also called the USDA Patented Strain, "is derived through nonrecombinant means, so you don't have to worry

SCOTT BAUER (K4390-31)



Soybean yields are higher after seed inoculation with the Nod+ nitrogen-fixing strain of bacteria. Developed by ARS, this inoculant has been rapidly growing in popularity.

about foreign genes in the bacterium's DNA," adds Kuykendall, with ARS' Molecular Plant Pathology Laboratory in Beltsville, Maryland.

Promising results from field studies also boosted acceptance by growers. For example, against *B. japonicum* I-110—a top soybean inoculant itself—the Nod+ strain formed 44 percent more nodules and fixed 50 percent more nitrogen. Generally speaking, a well-nodulated crop helps save on synthetic fertilizer costs and nourishes soils at rates less likely to affect groundwater, Hunter notes.

Large-scale testing of the Nod+ strain began shortly after Urbana began selling it in 1995. Based on those field trials, conducted by extension scientists at 377 sites in 18 states, the inoculant's use increased soybean yields by 2 to 3 bushels per acre.

In 1995, the first year of sales, Urbana inoculants containing the Nod+ strain were used on 220,000 acres of soybeans. Over 4 million acres' worth of inoculant was produced for 2001—a 20-fold increase. Since its introduction, Hunter estimates, the new inoculant has raised yields by nearly 30 million bushels. At \$5 per bushel, this means an additional \$150 million gross income for farmers.

In February, Hunter and Kuykendall received an ARS award for "superior effort" in transferring the inoculant technology to market.

Noting the 100-year history of soybean inoculants, Kuykendall comments that "the new strain's impact has been strong enough to show that we improved on an old, sustainable process and that what's good for the environment can actually make good economic sense as well."—By **Jan Suszkiw**, ARS.

This research is part of Plant Biological and Molecular Processes, an ARS National Program (#302) described on the World Wide Web at <http://www.nps.ars.usda.gov>.

To reach scientists mentioned in this article, contact Jan Suszkiw, USDA-ARS Information Staff, 5601 Sunnyside Ave., Beltsville, MD 20705; phone (301) 504-1630, fax (301) 504-1641, e-mail jsuszkiw@ars.usda.gov. ♦

High-Selenium Broccoli Stymies Some Cancers

Broccoli stores selenium in an especially useful form (called SeMSC) that is easily converted into an active anticancer agent. Now scientists have succeeded in greatly boosting the selenium in specially grown broccoli.

Earlier studies showed that laboratory rats fed experimental high-selenium broccoli and broccoli sprouts developed fewer precancerous lesions when exposed to known carcinogens than did rats given selenium salts—either selenate or selenite. The rats were fed the rough equivalent of a 200-microgram human dose of selenium daily.

The new tests showed that high-selenium broccoli sprouts protected the rats against precancerous lesions in the colon, while high-selenium broccoli protected against mammary tumors.

Specially produced for this research, the experimental broccoli heads and sprouts used in these studies aren't available commercially. And further study is needed to show whether these findings will also prove true in humans. *John W. Finley, USDA-ARS Grand Forks Human Nutrition Research Center, Grand Forks, North Dakota; phone (701) 795-8366, e-mail jfinley@gfhnrc.ars.usda.gov.*

Aromatic Compounds Suppress Potato Sprouts

Certain aromatic acids and jasmonates—compounds that impart the characteristic aroma of jasmine flowers—have been found to delay sprouting of stored potatoes if applied at the time of harvest. The aromatic acids and jasmonates could be especially useful for the premium organic potato market, which does not allow use of synthetic chemicals. The most widely applied sprout inhibitor registered for U.S. postharvest application to potatoes is synthetic.

Jasmonates are already used in the fragrance industry and as flavorings in foods. They have also been found to slightly improve the color of potatoes processed into chips and fries. Best of all, it takes just minute amounts to delay sprouting. The two treatments—jasmonates and aromatic acids—are patented and ready for commercial testing once a business partner is found to sign a patent license or cooperative agreement. *Edward C. Lulai, USDA-ARS Sugarbeet and Potato Research Laboratory, Fargo, North Dakota; phone (701) 239-1352, e-mail lulaie@fargo.ars.usda.gov.*

Waxy Wheat Cuts Bread Fat

A unique new kind of durum called waxy wheat has been found to function as its own shortening in bread recipes. Vegetable oil or another type of fat is often added to bread dough to improve crumb softness, loaf volume, and texture. Shortening also keeps bread from becoming stale too quickly.

This new kind of waxy durum wheat can replace vegetable shortening without losing desired properties of the bread. Not only would this save commercial bakers money, it would save consumers calories—about 26 grams of fat, or 234 calories per loaf.

Wheat is mostly starch, which is a polymer—or chain—of glucose molecules containing amylose (the straight-chain form) and amylopectin (the branched-chain form). Most wheat cultivars are about 24 percent amylose and 76 percent amylopectin. However, this new wheat contains an unusual type of starch that is 100 percent amylopectin. Researchers have been developing, evaluating, and testing applications for the new waxy durum wheat flour for about 5 years. *Douglas C. Doehlert, USDA-ARS Red River Valley Agricultural Research Center, Fargo, North Dakota; phone (701) 239-1413, e-mail doehlerd@fargo.ars.usda.gov.*

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